for human erythrocytes (*e.g.*, Guy Strain) and surrounds directly the growth of strains that are not hemolytic (*e.g.*, Oxford Strain). 2. Production of the red bands appears to depend upon a substance that diffuses through agar. 3. When sterile agar sections from plates previously inoculated with *S. aureus* are transferred onto blood plates, red bands develop around the transplanted section. 4. When plain agar is inoculated with the Guy Strain at a point near blood agar, a circular dense red band develops around the hemolytic zone in the blood medium; when inoculated with the Oxford Strain, a dense solid red band (half-moon) develops without hemolytic zone.

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Creatine Phosphorylkinase in Muscles of Dystrophic Mice.* (27310)

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Anomalies of creatine metabolism occur regularly in human muscular dystrophy, showing marked creatinuria(1) and diminished concentration of muscle creatine(2). Perkoff and Tyler(3) reported that the creatinuria of the Bar Harbor 129 strain of dystrophic mice is much less severe than human dystrophy. Kandutsch and Russell(4) showed that the decrease in muscle creatine is small in dystrophic mice when based on fat-free dry weight. This implies that certain biochemical dissimilarities exist between human dystrophy and the inherited dystrophy of mice.

From data obtained on biopsy specimens taken from gastrocnemius muscles of patients suffering from progressive muscular dystrophy, Ronzoni, Berg and Landau(5) found a close correlation between the low muscle creatine content and a depressed creatine-kinase activity. These investigators suggested that the low creatine content of dystrophic muscles is due to defective phosphorylation. However, Read and Nehorayan(6) showed that, in early vit. E-deficient rabbits, at a time when urinary excretion of creatine is rising, the creatine phosphorylating enzyme is actually in excess. Thus it would appear that increased urinary excretion of creatine and decreased muscle creatine may be due to causes other than depressed creatine phosphorylation.

The present report shows that there is no difference between the creatine phosphorylkinase activity from muscles of dystrophic mice and their littermate controls and is evidence of one further biochemical dissimilarity between human muscular dystrophy and that exhibited by the muscular dystrophic strain of mice.

Materials and methods. The mice studied were dystrophics (dy, dy) and heterozygous "carriers" (Dy, dy) obtained from the Roscoe B. Jackson Memorial Laboratory, Bar Harbor, Maine. The animals were 56 to 63 days old when used. Average weight of control animals was 24 g; that of dystrophic animals was 16 g.

The animals were given an intraperitoneal injection of sodium pentobarbital (40 mg/kg), the gastrocnemius muscles removed and chilled in chopped ice. Creatine phosphorylkinase was isolated from pooled muscle samples using the first 2 steps of procedure B of Kuby, Noda and Lardy(7). The enzyme was assayed using the procedure as outlined by Read and Nehorayan(6). Phosphorus was determined by the method of Fiske and Subbarow(8). Non-collagenous nitrogen (NCN) was extracted according to Lilienthal *et al.* (9). Nitrogen was determined by the method of Lanni, Dillon and Beard(10).

Results. In comparing the enzyme activi-

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TABLE I. Dry Weight, Fat-Free Dry Weight and Non-Collagenous Nitrogen of Muscles from Control and Dystrophic Mice.*

Animal	Dry wt, % of wet	Fat-free dry wt, % of wet	Non-collagenous nitrogen, mg/g wet wt
Control	23.8 ±.5†	19.8 ±.5	18.3 ± .6
Dystrophic	22.8 ±.2	14.0 ±.9	13.8 ± 1.0

* Figures represent avg of 9 animals.

† $\sqrt{\frac{Ed^2}{N(N-1)}}$.

ties between control and dystrophic muscle tissue, it was necessary to establish a proper reference base. In Table I are listed the values obtained for dry weight, fat-free dry weight and non-collagenous nitrogen from a series of control and dystrophic mice used in the present experiment. The figures show that the dry weights of control and dystrophic animals used were higher than those in the series analyzed by Weinstock, Epstein and Milhorat(11). The non-collagenous nitrogen was similar in the 2 control groups, but was lower in our dystrophic group than that of the dystrophic group analyzed by Weinstock *et al.* We have expressed the results of the present experiment in terms of the non-collagenous nitrogen.

Table II shows the values of the creatine phosphorylkinase activity determined for dystrophic and littermate controls. There was no difference between control and dystrophic mice when calculated on the basis of non-collagenous nitrogen. Table II also shows that administration of 0.2 mg of growth hormone (Somar, Armour Co) intraperitoneally for 14 days had no effect on creatine phosphorylkinase activity of the dystrophic muscle.

TABLE II. Creatine Phosphorylkinase Activity of Skeletal Muscle from Control and Dystrophic Mice.*

	Littermate control	Dystrophic	
	mM phosphorus transferred from ATP to creatine/g non-collagenous nitrogen		
Incubation time, min.	No treatment	No treatment	With growth hormone
2½	10.2 ±1.1†	10.2 ± .8	11.2 ± .7
5	17.5 ±1.2	18.5 ± .8	20.0 ± .9
10	27.5 ± .6	29.4 ±1.0	30.3 ±1.2

* Figures represent avg of determinations made on pooled samples from 10 groups of mice.

† $\sqrt{\frac{Ed^2}{N(N-1)}}$.

Discussion. Data on the creatine phosphorylating enzyme in human dystrophy(5) shows a good correlation with the depressed creatine content of the muscle and is offered as an explanation for the increased urinary excretion of creatine. The present report shows that the creatine phosphorylating enzyme in the dystrophic mouse is not different from non-dystrophic littermate controls. This may, in part, account for the small urinary excretion of creatine(3) and the less severe reduction in creatine content(4) of muscles from dystrophic mice when compared to similar determinations made on human muscular dystrophy.

Summary. The creatine phosphorylkinase activity of muscles taken from dystrophic mice is equal to that of non-dystrophic control mice.

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